PIKA HEATEX 36

For the rapid DNA isolation from enriched samples

Cat. No. 2034-1-1

Warning! Read the manual and the Safety Data Sheets before starting the analysis. Safety Data Sheets are available in the download area from www.pika-weihenstephan.de. All handling steps should be performed under sterile conditions. Wear appropriate protective clothing and powder-free gloves. The use of filter tips is recommended. For testing of Food and Environmental samples only.

Product description

The DNA Isolation Kit PIKA HeatEx 36 is intended for the extraction of DNA from bacteria and yeast cells in enriched samples and is optimized for Real time PCR. The microorganisms can either be isolated from solid surfaces i.e. colonies on membrane filters, or out of liquid samples. Prior isolation of single colonies is not necessary. Extracted DNA can directly be used for molecular analyses like PCR.

Kit content

Materials supplied are sufficient for 36 isolations.

Description	Amount	Storage*
Washing buffer A (yellow cap)	1 x 10.0 mL	4°C or room temperature
Lysis buffer B (blue cap)	1 x 10.0 mL	
Dilution buffer A	1 x 2,0 mL	

* Kit is shipped at ambient temperature

Materials required but not supplied

Material
Instruments and equipment
Benchtop microcentrifuge for 1.5 mL reaction tubes
Reaction tube mixer (Vortexer)
Thermoincubator or water bath set to 80°C
Pipettors
Consumables and reagents
Powder-free gloves
1.5 mL reaction tubes, safe-lock, sterile
Filter pipette tips

Procedural guidelines

1. Transfer the sample into a 1.5 mL reaction tube:
   a) Liquid samples:
   - 50 µL of a turbid, bacterial sample (previously enriched sample or spoiled product)
   - 1.0 – 1.5 mL of a clear sample (even larger sample sizes can be used)
   - 50 – 200 µL of yeast slurries to reach a pellet size of app. 2 mm in diameter after centrifugation (see fig. 1)

   b) *Colonies:* single colonies as well as different colonies can be processed together as one sample
   - Transfer 200 µL Washing buffer A and cell material into a 1.5 mL reaction tube. Skip steps 4. to 6.
2. Centrifuge for 3 min at 14,000 rpm (25,000 x g) or alternatively 10 min at 4,000 rpm (1,500 x g)
3. Control the pellet size. Pellet size should not exceed 2 mm in diameter (see fig. 1)
   If necessary, remove part of the pellet together with the liquid phase
4. Remove the liquid phase carefully and discard
5. Optional: Wash the pellet with 200 µL Washing buffer A,
   resuspend pellet and repeat steps 2 and 3
6. Add 200 µL of Lysis buffer B and resuspend by mixing briefly
7. Incubate sample at 80 °C ± 5 °C for 10 min in a thermoincubator or water bath
8. Centrifuge again as in step 2
   The pellet contains cell walls and other particles separated from the DNA
9. Transfer 100 µL of the liquid phase containing the DNA to a new 1.5 mL reaction tube and use the liquid phase for PCR. For long-term storage, freeze at -18 to –20 °C

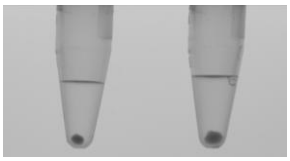

Fig. 1: recommended pellet sizes:
Left: max. pellet bacterial size
Right: max. pellet size for samples containing yeast

Dilution buffer A may be used to dilute the DNA prior to further analysis.

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